Resistance to black rot in a Spanish Brassica collection

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Introduction

Xanthomonas campestris pv. *campestris* (Xcc), causal agent of black rot, is widely distributed around the world in Brassica crops causing severe yield losses. The seedborne bacteria can survive in crop debris or crucifer weeds, introducing in the plant through hydathodes and wounds. While in warm and humid regions Xcc can cause plant dead, in coastal temperate areas it produces necrotic lesions on leaf margin, which decrease the value of the product on fresh market. In northwestern Spain, black rot has been recently identified in several Brassica crops (Lema et al. 2008). In this region, the production is mainly by small growers who do not use healthy plant material and disease-free seeds and, consequently, the pathogen can rapidly widespread. No studies involving either the pathogen or screens for resistance in this area have been conducted. For disease control the use of resistant cultivars is highly recommended but for most Brassica crops, especially in local crops, resistant cultivars are not offered and sources of resistance are very limited or even unknown. In the last years, the search for new sources of resistance has been race-specific since the existence of six races of the pathogen was described by Vicente et al. (2001). In addition to monogenic race-specific, a quantitative race-nonspecific resistance has been described (Soengas et al. 2007, Taylor et al. 2002). Recently, Fargier and Manceau (2007) added three new races (7 to 9), being races 1 and 4 are the most virulent and widespread, accounting for most black rot cases around the world. Sources of resistance to Xcc in Brassica genomes have been examined by different researchers but the use of resistant cultivars has only had limited success and available sources with useful levels of resistance are scarce. Moreover, most of these works focused in cabbage due to its economical value, while the search for resistance in other Brassica crops has been more restricted. Therefore, the objective of this work was to identify new sources of resistance to races 1 and 4 of Xcc in several Brassica crops.

Materials and Methods

Five hundred and twenty six accessions from Germplasm Bank in the Misión Biológica de Galicia-CSIC (Spain) belonging to three *Brassica* species were screened for black rot resistance together with several resistant and susceptible controls establish by Vicente et al. (2001).These accessions comprises 76 of *B. napus* (including *napus, napobrassica, oleifera* and *pabularia* groups), 256 of *B. oleracea* (including *acephala, capitata* and *costata* groups) and 194 of *B. rapa* subsp. *rapa*. Bacterial isolates of race 1 type strain HRI3811and race 4 type strain HRI1279A provided by Warwick-HRI-Wellesbourne, UK were used. Bacterial cultures were grown in bacterial screening media 523 at 30 ºC during 48 h prior to inoculation and were diluted in sterile tap water until suspension reached a density of 5×10^{8} cfu mL^{-1}. Fourteen to sixteen four weeks old plants per accession were artificially inoculated in greenhouse conditions following the methodology described by Lema-Marquez et al. (2007). The disease severity caused by each Xcc race was quantitatively rated using a 1 (resistant) to 9 (susceptible) scale. Analyses of variance were performed for disease score and were combined across races by using the GLM procedure of SAS (2002). Accessions and races were considered as fixed effects whereas replications (plants within accessions) were considered as a random factor. Comparisons of means were performed for each trait by using Fisher's protected Least Significant Difference (LSD) at the 0.05 level of probability. The sums of squares for accessions were orthogonally divided into groups in *B. napus* and *B. oleracea* species.

Results and Discussion

Brassica napus

Race 1 was more virulent on the tested materials than race 4. No race-specific resistance was found to race 1. Most cultivars were susceptible except Russian kale, from the *pabularia* group, which showed some resistant plants and some other accessions with some partially resistant plants. High levels of race-specific resistance to race 4 were found, particularly in the *pabularia* group, although great variability within accessions was identified. Three improved cultivars (Ragged Jack kale, Friese Gele, Valle del Oro) and four landraces (Russian kale, MBG-BRS0037, MBG-BRS0041, MBG-BRS0131) showed plants with some degree of resistance to both races (Table 1), then possible race-nonspecific resistance can be involved. These accessions could be directly used in breeding programs, either to improve the cultivar per se or like donors of race-specific resistance to other Brassica cultivars. Different selection criteria applied on *B. napus* crops according to their use could lead to an indirect selection for Xcc resistance.

Brassica oleracea

The accessions performed statistically distinct against two races, being race 1 slightly more virulent on tested materials than race 4. Most accessions were susceptible to both races, except cabbage cultivars ‘Balón’ and ‘Quintal de Alsacia’ showing some plants with different level of resistance to races 1 and 4, indicating that race-nonspecific resistance can be involved. Kale landraces MBG-BRS0286 and MBG-BRS0070 showed an intermediate mean disease score for races 1 and 4, respectively (Table 1). These accessions can be crossed to cabbage cultivars and may provide new combinations of resistance genes with protection against black rot in cabbage production areas.

Brassica rapa

Partial resistance was found in several landraces to race 1 and resistance and partial resistance in several landraces to race 4. Three landraces were identified as potential race-nonspecific resistant (Table 1). Sources of resistance were identified in different crops of the subspecies (turnips, turnip greens and turnip tops) and they can be grown directly after selection for resistance or they can be used to introgress resistance in other germplasm or commercial varieties.

Conclusions

Race 1 was much more virulent on tested materials that race 4. Most of the *B. rapa* (72%) and *B. napus* (55%) accessions showed resistance to race 4 although a great variability within accessions was found, probably due to a mixture of genotypes. Noteworthy were data recorded in *B. oleracea* accessions where existing sources of resistance are limited. According these results local materials can be used in breeding programs taking in account that they are heterogeneous due to mixture of genotypes and intercrossing among varieties probably associated with poor isolation.

References

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Table 1. Percentage of resistant plants and mean disease score for Xcc races 1 and 4 for the most promising accessions from Germplasm Bank in the Misión Biológica de Galicia included in this study

Species	Group	Accession	Resistant plants		Mean disease score	
			Race 1	Race 4	Race 1	Race 4
			---------(%)--------		---------(1-9)---------	
Brassica napus	*pabularia*	MBG-BRS0041	0	100	8.6	1.8
	pabularia	Russian kale	25	50	5.3	3.5
	oleifera	Valle del Oro	0	47	8.6	3.9
	napobrassica	Friese Gele	0	33	7.1	5.3
	pabularia	MBG-BRS0037	0	33	8.1	5.9
	pabularia	MBG-BRS0131	0	33	7.8	5.9
	napus	Ragged Jack kale	0	12	7.9	7.2
Brassica oleracea	*acephala*	MBG-BRS0070	0	63	9.0	4.5
	capitata	Balon	7	8	6.8	7.1
	capitata	Quintal de Alsacia	22	0	6.7	7.9
	acephala	MBG-BRS0286	0	0	6.5	9.0
Brassica rapa		MBG-BRS0259	0	100	8.3	1.3
		MBG-BRS0417	0	53	6.9	3.9
		MBG-BRS0262	0	42	7.7	4.2
		MBG-BRS0215	20	17	6.2	5.3
		MBG-BRS0479	0	66	6.7	6.6